## Validation data for 293-hMyD88 cells

https://www.invivogen.com/293-hmyd88

## For research use only

Version 21E04-ED

293-hMyD88 cells were generated from the human embryonic kidney (HEK)-293 cell line, through the stable and constitutive expression of the human (h)MyD88 gene. 293-hMyD88 cells have been specifically designed for use in InvivoGen's assay to study SARS-CoV-2 Spike-ACE2-dependent cell fusion, which relies on the transfer of MyD88 from the 'donor cell line' to a permissive 'acceptor cell line' expressing an inducible NF-KB-SEAP reporter gene (Figure 1).

## Assessing cell fusion with InvivoGen's COVID-19 cell lines

To generate the 'donor cell line', 293-hMyD88 cells were transiently transfected with InvivoGen's pUNO1-Spike expression plasmid, which encodes the Wuhan-Hu-1 Spike with a functional furin cleavage site to faciliate cell fusion. Upon co-culture of a dilution series of the 'donor cell line' with the acceptor cell line, HEK-Blue™ hACE2, cell fusion was triggered, and MyD88 activated a signalling cascade in the 'acceptor cell line', ultimately leading to readily assessable SEAP production.

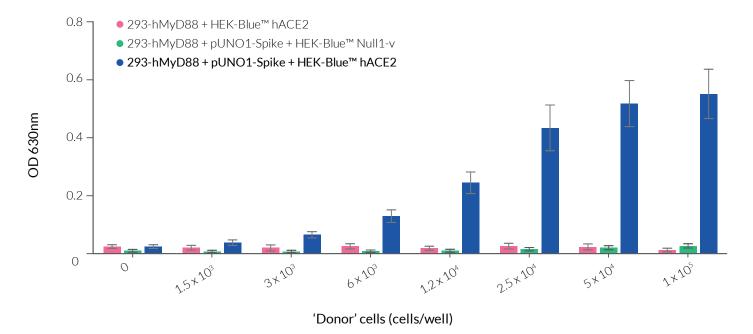


Figure 1: Assessing cell fusion with 293-hMyD88 cells. 293-hMyD88 cells were transiently transfected with a SARS-CoV-2 Spike expression plasmid (pUNO1-Spike) using LyoVec<sup> $\top$ </sup>. After 24 hours, the cells were washed, and a dilution series of the 'donor' non transfected or the 293-hMyD88-Spike cells was co-cultured with either  $2.0 \times 10^4$  HEK-Blue<sup> $\top$ </sup> Null1-v or HEK-Blue<sup> $\top$ </sup> hACE2 cells. After overnight incubation, cell fusion was assessed by measuring the activity of SEAP in the supernatant using QUANTI-Blue<sup> $\top$ </sup> Solution, a SEAP detection reagent. Data are presented as OD630nm  $\pm$  SEM.



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